SEQUENCE LISTING

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<110> WHITELAW, CHRISTOPHER BRUCE ALEXANDER
      CLARK, ANTHONY JOHN
      WOLF, CHARLES ROLAND
<120> MULTI-REPORTER GENE MODEL FOR TOXICOLOGICAL SCREENING
<130> 102286.155 US1
<140> 10/522,356
<141> 2005-01-26
<150> PCT/GB03/003192
<151> 2003-07-25
<150> GB 0217402.7
<151> 2002-07-26
<160> 41
<170> PatentIn version 3.3
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<213> Unknown
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Leu

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gaa tat tot gtg acg tat gat gga tto aat aca ttt act ata cot aag Glu Tyr Ser Val Thr Tyr Asp Gly Phe Asn Thr Phe Thr Ile Pro Lys 100 105 110	336
aca gac tat gat aac ttt ctt atg gct cat ctc att aac gaa aag gat Thr Asp Tyr Asp Asn Phe Leu Met Ala His Leu Ile Asn Glu Lys Asp 115 120 125	384
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Met Lys Met Leu Leu Leu Cys Leu Gly Leu Thr Leu Val Cys Val 1 5 15 15 15 His Ala Glu Glu Ala Ser Ser Thr Gly Arg Asn Phe Asn Val Glu Lys 25 30 Ile Asn Gly Glu Trp His Thr Ile Ile Leu Ala Ser Asp Lys Arg Glu	
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<211> 179

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic recombinant mMUP reporter molecule

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Thr Met Glu Ala Ser Ser Thr Gly Arg Asn Phe Asn Val Glu Lys Ile 20 25 30

Asn Gly Glu Trp His Thr Ile Ile Leu Ala Ser Asp Lys Arg Glu Lys 35 40 45

Ile Glu Asp Asn Gly Asn Phe Arg Leu Phe Leu Glu Gln Ile His Val 50 55 60

Leu Glu Lys Ser Leu Val Leu Lys Phe His Thr Val Arg Asp Glu Glu 65 70 75 80

Cys Ser Glu Leu Ser Met Val Ala Asp Lys Thr Glu Lys Ala Gly Glu 85 90 95

Tyr	Ser	Val	Thr 100	Tyr	Asp	Gly	Phe	Asn 105	Thr	Phe	Thr	Ile	Pro 110	Lys	Thr	
Asp	Tyr	Asp 115	Asn	Phe	Leu	Met	Ala 120	His	Leu	Ile	Asn	Glu 125	Lys	Asp	Gly	
Glu	Thr 130	Phe	Gln	Leu	Met	Gly 135	Leu	Tyr	Gly	Arg	Glu 140	Pro	Asp	Leu	Ser	
Ser 145	Asp	Ile	Lys	Glu	Arg 150	Phe	Ala	Gln	Leu	Cys 155	Glu	Lys	His	Gly	Ile 160	
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tac Tyr	gtg Val 50	gag Glu	gag Glu	ctg Leu	aag Lys	ccc Pro 55	acc Thr	ccc Pro	gag Glu	ggc Gly	aac Asn 60	ctg Leu	gag Glu	atc Ile	ctg Leu	192

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	aaa Lys	_		 _	_	_			_		_		336
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<211> 179

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic recombinant BLGm reporter molecule

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Leu Asp Ile Gln Lys Val Ala Gly Thr Trp His Ser Leu Ala Met Ala 20 25 30

Ala Ser Asp Ile Ser Leu Leu Asp Ala Gln Ser Ala Pro Leu Arg Val 35 40 45

Tyr Val Glu Glu Leu Lys Pro Thr Pro Glu Gly Asn Leu Glu Ile Leu
50 60

Leu Gln Lys Trp Glu Asn Gly Glu Cys Ala Gln Lys Lys Ile Ile Ala 65 70 75 80

Glu Lys Thr Lys Ile Pro Ala Val Phe Lys Ile Asp Ala Leu Asn Glu 85 90 95

Asn Lys Val Leu Val Leu Asp Thr Asp Tyr Lys Lys Tyr Leu Leu Phe 100 105 110

Cys Met Glu Asn Ser Ala Glu Pro Glu Gln Ser Leu Ala Cys Gln Cys 115 120 125

Leu Val Arg Thr Pro Glu Val Asp Asn Glu Ala Leu Glu Lys Phe Asp 130 135 140

Lys Ala Leu Lys Ala Leu Pro Met His Ile Arg Leu Ala Phe Asn Pro 145 150 155 160

Thr Gln Leu Glu Gly Gln Cys His Val Glu Gln Lys Leu Ile Ser Glu 165 170 175

Glu Asp Leu

<210> 19

<211> 214

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic modified MUP protein produced from the pSecTag vector

<400> 19

Met Glu Thr Asp Thr Leu Leu Leu Trp Val Leu Leu Leu Trp Val Pro 1 5 10 15

Gly Ser Thr Gly Asp Ala Ala Gln Pro Ala Arg Arg Ala Arg Thr 20 25 30

Lys Leu Gly Thr Glu Leu Gly Ser Met Glu Gln Lys Leu Ile Ser Glu 35 40 45

Glu Asp Leu Thr Met Glu Ala Ser Ser Thr Gly Arg Asn Phe Asn Val $50 \\ \hspace{1.5cm} 55 \\ \hspace{1.5cm} 60$

Glu Lys Ile Asn Gly Glu Trp His Thr Ile Ile Leu Ala Ser Asp Lys 65 70 75 80

Arg Glu Lys Ile Glu Asp Asn Gly Asn Phe Arg Leu Phe Leu Glu Gln 85 90 95

Ile His Val Leu Glu Lys Ser Leu Val Leu Lys Phe His Thr Val Arg
100 105 110

Asp Glu Glu Cys Ser Glu Leu Ser Met Val Ala Asp Lys Thr Glu Lys 115 120 125

Ala Gly Glu Tyr Ser Val Thr Tyr Asp Gly Phe Asn Thr Phe Thr Ile 130 135 140

Pro Lys Thr Asp Tyr Asp Asn Phe Leu Met Ala His Leu Ile Asn Glu 145 150 155

Lys Asp Gly Glu Thr Phe Gln Leu Met Gly Leu Tyr Gly Arg Glu Pro \$165\$ \$170\$ \$175\$

Asp Leu Ser Ser Asp Ile Lys Glu Arg Phe Ala Gln Leu Cys Glu Lys
180 185 190

His Gly Ile Leu Arg Glu Asn Ile Ile Asp Leu Ser Asn Ala Asn Arg 195 200 205

Cys Leu Gln Ala Arg Glu 210

<210> 20

<211> 243

<212> PRT

<213> Artificial Sequence

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<223> Description of Artificial Sequence: Synthetic modified MUP protein produced from the pSecTag vector

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Cys Leu Gly Leu Thr Leu Val Cys Val His Ala Glu Glu Ala Ser Ser 35 40 45

Thr Gly Arg Asn Phe Asn Val Glu Lys Ile Asn Gly Glu Trp His Thr 50 55 60

Ile Ile Leu Ala Ser Asp Lys Arg Glu Lys Ile Glu Asp Asn Gly Asn 65 70 75 80

Phe Arg Leu Phe Leu Glu Gln Ile His Val Leu Glu Lys Ser Leu Val 85 90 95

Leu Lys Phe His Thr Val Arg Asp Glu Glu Cys Ser Glu Leu Ser Met 100 105 110

Val Ala Asp Lys Thr Glu Lys Ala Gly Glu Tyr Ser Val Thr Tyr Asp 115 120 125

Gly Phe Asn Thr Phe Thr Ile Pro Lys Thr Asp Tyr Asp Asn Phe Leu 130 140

Met Ala His Leu Ile Asn Glu Lys Asp Gly Glu Thr Phe Gln Leu Met 145 150 155 160

Gly Leu Tyr Gly Arg Glu Pro Asp Leu Ser Ser Asp Ile Lys Glu Arg 165 170 175

Phe Ala Gln Leu Cys Glu Lys His Gly Ile Leu Arg Glu Asn Ile Ile 180 185 190

Asp Leu Ser Asn Ala Asn Arg Cys Leu Gln Ala Arg Glu Gln Lys 195 200 205

Leu Ile Ser Glu Glu Asp Leu Ala Ala Ala Arg Gly Gly Pro Glu Gln 210 215 220

Lys Leu Ile Ser Glu Glu Asp Leu Asn Ser Ala Val Asp His His His 225 230 235

His His His

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic modified MUP protein produced from the pSecTag vector

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Gly Ser Thr Gly Asp Ala Ala Gln Pro Ala Lys Met Leu Leu Leu 20 25 30

Cys Leu Gly Leu Thr Leu Val Cys Val His Ala Glu Glu Ala Ser Ser 35 40 45

Thr Gly Arg Asn Phe Asn Val Glu Lys Ile Asn Gly Glu Trp His Thr 50 55 60

Ile Ile Leu Ala Ser Asp Lys Arg Glu Lys Ile Glu Asp Asn Gly Asn 65 70 75 80

Phe Arg Leu Phe Leu Glu Gln Ile His Val Leu Glu Lys Ser Leu Val 85 90 95

Leu Lys Phe His Thr Val Arg Asp Glu Glu Cys Ser Glu Leu Ser Met
100 105 110

Val Ala Asp Lys Thr Glu Lys Ala Gly Glu Tyr Ser Val Thr Tyr Asp 115 120 125

Gly Phe Asn Thr Phe Thr Ile Pro Lys Thr Asp Tyr Asp Lys Leu Gly 130 135 140

Glu Lys Asp Gly Glu Thr Phe Gln Leu Met Gly Leu Tyr Gly Arg Glu 165 170 175 Pro Asp Leu Ser Ser Asp Ile Lys Glu Arg Phe Ala Gln Leu Cys Glu 180 185 190

Lys His Gly Ile Leu Arg Glu Asn Ile Ile Asp Leu Ser Asn Ala Asn 195 200 205

Arg Cys Leu Gln Ala Arg Glu Glu Gln Lys Leu Ile Ser Glu Glu Asp 210 215 220

Leu Ala Ala Arg Gly Gly Pro Glu Gln Lys Leu Ile Ser Glu Glu 225 230 235 240

Asp Leu Asn Ser Ala Val Asp His His His His His Essen 245 250

<210> 22

<211> 259

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic modified MUP protein produced from the pSecTag vector

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Gly Ser Thr Gly Asp Ala Ala Gln Pro Ala Lys Met Leu Leu Leu 20 25 30

Cys Leu Gly Leu Thr Leu Val Cys Val His Ala Glu Glu Ala Ser Ser 35 40 45

Thr Gly Arg Asn Phe Asn Val Glu Lys Ile Asn Gly Glu Trp His Thr 50 55 60

Ile Ile Leu Ala Ser Asp Lys Arg Glu Lys Ile Glu Asp Asn Gly Asn 65 70 75 80

Phe Arg Leu Phe Leu Glu Gln Ile His Val Leu Glu Lys Ser Leu Val 85 90 95

Leu Lys Phe His Thr Val Arg Asp Glu Glu Cys Ser Glu Leu Ser Met 100 105 110

Val A	Ala	115	гÀг	THE	GIU	rys	120	GIĄ	GIU	Tyr	ser	125	Inr	Tyr	Asp	
Gly F	Phe 130	Asn	Thr	Phe	Thr	Ile 135	Pro	Lys	Thr	Asp	Tyr 140	Asp	Lys	Leu	Asn	
Val A 145	Arg	Phe	Ser	Thr	Ile 150	Val	Arg	Arg	Arg	Ala 155	Glu	Phe	Asn	Phe	Leu 160	
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Asp I	Leu 210	Ser	Asn	Ala	Asn	Arg 215	Cys	Leu	Gln	Ala	Arg 220	Glu	Glu	Gln	Lys	
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<212> PRT

<213> Ovis aries

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Val Ala Gly Thr Trp His Ser Leu Ala Met Ala Ala Ser Asp Ile Ser 35 40 45

Leu Leu Asp Ala Gln Ser Ala Pro Leu Arg Val Tyr Val Glu Glu Leu 50 55 60

Lys Pro Thr Pro Glu Gly Asn Leu Glu Ile Leu Leu Gln Lys Trp Glu 65 70 75 80

Asn Gly Glu Cys Ala Gln Lys Lys Ile Ile Ala Glu Lys Thr Lys Ile 85 90 95

Pro Ala Val Phe Lys Ile Asp Ala Leu Asn Glu Asn Lys Val Leu Val

Leu Asp Thr Asp Tyr Lys Lys Tyr Leu Leu Phe Cys Met Glu Asn Ser 115 120 125

Ala Glu Pro Glu Gln Ser Leu Ala Cys Gln Cys Leu Val Arg Thr Pro 130 \$135\$

Leu Pro Met His Ile Arg Leu Ala Phe Asn Pro Thr Gln Leu Glu Gly 165 170 175

Gln Cys His Val

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Ile Asn Gly Glu Trp His Thr Ile Ile Leu Ala Ser Asp Lys Arg Glu

40

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Glu	Cys	Ser	Glu	Leu 85	Ser	Met	Val	Ala	Asp 90	Lys	Thr	Glu	Lys	Ala 95	Gly	
Glu	Tyr	Ser	Val 100	Thr	Tyr	Asp	Gly	Phe 105	Asn	Thr	Phe	Thr	Ile 110	Pro	Lys	
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<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Synthetic
 GST coding sequence derived from pGEX6p-1

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<210> 29

<211> 687

<212> DNA

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 GST coding sequence derived from pGEX6p-1

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	ctt ctt Leu Leu 20												96
	cgc gat Arg Asp 35												144
ggt ttg Gly Leu 50	gag ttt Glu Phe							_		_	_		. 192
tta aca Leu Thr 65	cag tct Gln Ser	atg gc Met Ala 70	atc a Ile	ata Ile	cgt Arg	tat Tyr	ata Ile 75	gct Ala	gac Asp	aag Lys	cac His	aac Asn 80	240
atg ttg Met Leu		_		_	_	_				_		_	288
gga gcg Gly Ala		Asp Ile											336
aaa gac Lys Asp			ı Lys										384
atg ctg Met Leu 130													432
ggt gat Gly Asp 145			Pro										480
gtt gtt Val Val													528
gtt tgt Val Cys													576
ttg aaa Leu Lys	tcc agc Ser Ser 195	aag tat Lys Ty	· Ile .	gca Ala 200	tgg Trp	cct Pro	ttg Leu	cag Gln	ggc Gly 205	tgg Trp	caa Gln	gcc Ala	624

acg ttt ggt ggt ggc gac cat cct cca aaa tcg gat ctg gaa gtt ctg 672 Thr Phe Gly Gly Asp His Pro Pro Lys Ser Asp Leu Glu Val Leu 215 220 687 ttc cag ggg ccc ctg Phe Gln Gly Pro Leu 225 <210> 30 <211> 229 <212> PRT <213> Artificial Sequence <220> <223> Description of Artificial Sequence: Synthetic GST coding sequence derived from pGEX6p-1 <400> 30 Met Ser Pro Ile Leu Gly Tyr Trp Lys Ile Lys Gly Leu Val Gln Pro 10 Thr Arg Leu Leu Glu Tyr Leu Glu Glu Lys Tyr Glu Glu His Leu Tyr Glu Arg Asp Glu Gly Asp Lys Trp Arg Asn Lys Lys Phe Glu Leu 35 40 45 Gly Leu Glu Phe Pro Asn Leu Pro Tyr Tyr Ile Asp Gly Asp Val Lys 50 55 60 Leu Thr Gln Ser Met Ala Ile Ile Arg Tyr Ile Ala Asp Lys His Asn 65 70 75 Met Leu Gly Gly Cys Pro Lys Glu Arg Ala Glu Ile Ser Met Leu Glu Gly Ala Val Leu Asp Ile Arg Tyr Gly Val Ser Arg Ile Ala Tyr Ser Lys Asp Phe Glu Thr Leu Lys Val Asp Phe Leu Ser Lys Leu Pro Glu 115 120 125 Met Leu Lys Met Phe Glu Asp Arg Leu Cys His Lys Thr Tyr Leu Asn 135 Gly Asp His Val Thr His Pro Asp Phe Met Leu Tyr Asp Ala Leu Asp

155

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Val Val Leu Tyr Met Asp Pro Met Cys Leu Asp Ala Phe Pro Lys Leu
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Val Cys Phe Lys Lys Arg Ile Glu Ala Ile Pro Gln Ile Asp Lys Tyr
                                 185
Leu Lys Ser Ser Lys Tyr Ile Ala Trp Pro Leu Gln Gly Trp Gln Ala
Thr Phe Gly Gly Asp His Pro Pro Lys Ser Asp Leu Glu Val Leu
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                                            220
Phe Gln Gly Pro Leu
<210> 31
<211> 24
<212> DNA
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Synthetic
      protease cleavage site
<220>
<221> CDS
<222> (1)..(24)
<400> 31
ctg gaa gtt ctg ttc cag ggg ccc
                                                                       24
Leu Glu Val Leu Phe Gln Gly Pro
<210> 32
<211> 8
<212> PRT
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Synthetic
      protease cleavage site
<400> 32
Leu Glu Val Leu Phe Gln Gly Pro
                5
<210> 33
<211> 32
<212> DNA
<213> Artificial Sequence
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<220>
<223> Description of Artificial Sequence: Synthetic
      nucleotide sequence that contains a Kozak signal,
      start codon and NcoI-KpnI-XbaI-PstI linker
<400> 33
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                                                                        32
<210> 34
<211> 21
<212> DNA
<213> Artificial Sequence
<223> Description of Artificial Sequence: Synthetic
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      AvrII-ApaI-SbfI linker
<400> 34
tgcctagggc cctgcagggt a
                                                                        21
<210> 35
<211> 31
<212> DNA
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Synthetic
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actagtgaat tcatgcattg agctagccat c
                                                                        31
<210> 36
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<222> (5)
<223> a, g, c, t, unknown or other
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<221> modified_base
<222> (6)..(8)
<223> a, g, c, t, unknown or other and see
      specification for further embodiments
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<220>
<221> modified_base
<222> (9)
<223> a, g, c, t, unknown or other
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                                                                        13
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<210> 37
<211> 18
<212> DNA
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Synthetic
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<400> 37
aagcttggaa ccggatcc
                                                                        18
<210> 38
<211> 18
<212> DNA
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Synthetic
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      restriction sites
<400> 38
ggatcctctt cagaattc
                                                                        18
<210> 39
<211> 39
<212> DNA
<213> Artificial Sequence
<223> Description of Artificial Sequence: Synthetic
      nucleotide sequence that contains c-myc epitope
      tag, stop codon and NheI restriction site
<400> 39
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<210> 40
<211> 9
<212> DNA
<213> Artificial Sequence
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<220>
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<223> a, c, g, t, unknown or other and see
      specification for further embodiments
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